



# Comparison of four aquatic plant treatment systems for nutrient removal from eutrophied water



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## HIGHLIGHTS

- We compare nutrient removal behaviors of four aquatic plant treatment systems.
- The kinetics of nutrient uptake rely on nutrient forms and plant species.
- We report the main pathways of nutrient removal in the plant treatment system.

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## ABSTRACT

Nutrient removal behaviors of four aquatic plant treatment systems (*Oenanthe javanica*, *Iris pseudacorus* L., *Canna lily*, and *Potamogeton crispus*) were systematically examined and compared. The kinetics of nutrient uptake were conducted with the standard depletion method. All four aquatic species exhibited a strong preference of ammonium nitrogen ( $\text{NH}_4^+\text{-N}$ ) over nitrate nitrogen ( $\text{NO}_3^-\text{-N}$ ) and nitrite nitrogen ( $\text{NO}_2^-\text{-N}$ ). Main pathways of nutrient removal in the aquatic plant treatment system were examined in details. It was estimated that direct assimilation by plants accounted for 28.2–34.5% of N reduction and 25.2–33.4% of P reduction while substrate absorption accounted for 7.2–25.5% of N reduction and 7.3–25.0% of P reduction. The activity of urease and phosphatase in the substrates could indicate the aquatic plant treatment system's capability for reducing TN and soluble P load.

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## 1. Introduction

Harmful algal blooms caused by eutrophication have threatened not only drinking water safety but also the ecological integrity of aquatic environment and the sustainability of socio-economic development around the world (Paerl et al., 2011). Excessive anthropogenic nutrient discharge is the main cause of eutrophication and the subsequent proliferation and spread of hazardous algae (Anderson et al., 2008). Nitrogen (N) and phosphorous (P) have been identified as the two main nutrients needed to control for mitigating the serious situation of eutrophication. Stringent discharge standards have been stipulated to control the discharge of the two nutrients into the aquatic environment (Conley et al., 2009; USEPA, 2012). However, the alleviation of algae bloom and the improvement of water quality in eutrophied water bodies could not be achieved solely through reducing external nutrient loads, since internal nutrient release could also contribute signifi-

cantly to eutrophication (Guo et al., 2014). Hence, a variety of technologies for cutting internal nutrient load and facilitating ecosystem recovery have been developed and implemented (Zhang et al., 2014), including the mechanical removal of algae (Lam et al., 1995), the application of algacide, the dredging of lake sediments (Kleeberg and Kohl, 1999), and the cultivation of aquatic plants for nutrient removal and ecosystem recovery (Dai et al., 2012). With the advantages of low cost and environmental friendliness, aquatic plant treatment systems have been increasingly recognized as a useful approach for controlling eutrophication. In such systems, nutrients were removed simultaneously by plants and the diverse microbiological communities that depend on the roots of the aquatic plants to thrive.

Previous studies of nutrient removal by aquatic plants have been mostly limited to constructed wetland systems (Zimmels et al., 2008; Gao et al., 2014). Breen (1990) found that the constructed wetland system was effective in removing nutrients with a 51% N removal rate and 67% P removal rate. They concluded that plant biomass was the major nutrient storage repository. Huett et al. (2005) treated agricultural runoff with *Phragmites australis*, and reduced its N and P load by 76% and 86%, respectively. Overall,

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the performance of the constructed wetland system in nutrient removal could be affected by a variety of operating conditions such as temperature, hydraulic residence time, and plant coverage (Zhang et al., 2012; Peng et al., 2014).

Previous research has indicated that introduction of aquatic plants into eutrophied water bodies could facilitate a long-term improvement in water quality (Coveney et al., 2002; Hu et al., 2010). Qiu et al. (2001), for example, observed that water quality was remarkably improved when *Potamogeton maackianus* was introduced into hypertrophic lakes. In shallow eutrophied water bodies, aquatic plants can assimilate a large amount of N and P from sediments via their roots during the growing season (Xie et al., 2013). In addition, the roots of aquatic plants could create a variety of microenvironments for microorganisms to breed and reproduce. However, compared to constructed wetlands, fewer studies have been conducted to systematically investigate the behaviors of the major components of the aquatic plant treatment systems for nutrient removal in eutrophied water bodies where residence time is no longer an influencing factor. In this study, the nutrient removal behaviors of four aquatic plant treatment systems (*Oenanthe javanica* (OEJ), *Iris pseudacorus* L. (IRP), *Canna lily* (CAL), and *Potamogeton crispus* (POC)) in eutrophied water bodies were systematically examined and compared. First, the standard depletion method was used to examine and compare the kinetics of nutrient uptake by the four aquatic plant species. Second, the change of nutrient contents in the solution, plant, and substrate components of each aquatic plant treatment systems was determined to evaluate each component's contribution to nutrient reduction. Finally, regression relationships between substrate enzyme activity and nutrient removal rates were estimated and compared among the four aquatic plant treatment systems. Insights into the above mechanisms and behaviors of nutrient removal by aquatic plant treatment systems facilitate the design and implementation of effective aquatic plant treatment systems for eutrophication mitigation.

## 2. Methods

### 2.1. Kinetics of nutrient uptake by aquatic plants

Hoagland nutrient solution was prepared except without N or P. Its pH was adjusted to 6.0 to avoid ammonia volatilization. Ammonium sulfate, sodium nitrate, sodium nitrite, and sodium dihydrogen phosphate were then added to provide  $\text{NH}_4^+\text{-N}$ ,  $\text{NO}_3^-\text{-N}$ ,  $\text{NO}_2^-\text{-N}$ , and orthophosphate ( $\text{PO}_4^{3-}$ ) to the solution, respectively. Both N and P containing compounds were added at the gradient of 0.1, 0.5, 1.0, 2.0, 4.0, 10.0, 15.0, and 20.0  $\text{mg L}^{-1}$ .

Nutrient uptake rates by the four aquatic species were studied with the standard depletion method. Plants of the same weight were used in each treatment due to their variable size. 25 g of the seedlings were used in the treatments with OEJ, IRP, and CAL, while 8 g were used in those with POC because of its being a submerging species. After a starvation period of 20 h, seedlings were rinsed with de-ionized water, dried with absorbent paper, and transplanted into sponge sheets placed in the 500 mL beakers containing 300 mL nutrient solution of various N and P concentrations.

The nutrient uptake experiments were conducted in dark at a temperature of  $25 \pm 2$  °C and a relative humidity of 75%. After 6 h, the plants were taken out and dried with absorbent paper. The roots of OEJ, IRP, and CAL were then separated, and dried at 70 °C for 72 h to determine their dry weight. The whole POC was dried and weighted in the same manner.

N and P contents of the roots of the plants were measured before and after each treatment, and used to calculate the total amount of nutrient uptake as well as the average uptake rate

during each treatment. Absorption kinetics parameters such as  $V_{\text{max}}$  and  $K_m$  were calculated through fitting the Michaelis–Menten equation and the Hofstee transformation.  $V_{\text{max}}$  relates mainly to the number of ion transporters in cell membranes, and could reflect the uptake potential for the ion.  $K_m$  reflects the affinity of the transporter to the ion, with higher affinity indicating higher absorption efficiency. The two parameters allowed to make comparisons of nutrient uptake efficiencies among different plant species, as well as to provide insights into the plants' nutrient uptake mechanisms (Zhou et al., 2011).

### 2.2. Distribution and transport of nutrients in the aquatic plant treatment systems

Quartz sand was washed with de-ionized water and spread evenly over the bottom of the 5 L beakers at a depth of 10 cm. Artificial eutrophic solution prepared with tap water, glucose ( $\text{C}_6\text{H}_{12}\text{O}_6$ ), urea ( $\text{CO}(\text{NH}_2)_2$ ), and  $\text{KH}_2\text{PO}_4$  at an initial N and P concentration of 20 mg/L and 2 mg/L was added slowly to the beakers till their top. Healthy plants of the same wet weight were planted in the beakers under the natural sunlight. No plants were planted in the control group. Water quality parameters, such as pH, redox potential (ORP), TN,  $\text{NH}_4^+\text{-N}$ ,  $\text{NO}_3^-\text{-N}$ ,  $\text{NO}_2^-\text{-N}$ , TP, and  $\text{PO}_4^{3-}\text{-P}$ , were measured 1 day after the beginning of the experiment and every 6 days afterwards for 25 days. Physiological indicators of plant growth such as biomass and N and P contents of plant organs were measured at the start and finish of the experiments.

### 2.3. Analysis methods

#### 2.3.1. Analysis of water samples

Water quality parameters were measured following the guidelines set by Chinese Ministry of Environmental Protection (Chinese Environmental Protection Bureau, 2002). TP and  $\text{PO}_4^{3-}\text{-P}$  were measured with the ammonium molybdate spectrophotometric method.  $\text{NH}_4^+\text{-N}$  was determined by the method of Nessler's reagent photometry. TN was determined using Alkaline potassium persulfate digestion, followed by UV spectrophotometric method as for  $\text{NO}_3^-\text{-N}$ .  $\text{NO}_2^-\text{-N}$  was measured with the cadmium reduction method. pH and ORP were determined by the method of glass electrode method and electrometric method, respectively.

#### 2.3.2. Analysis of plant samples

After the experiments, the plants were removed completely from the solution and substrates entangled in the roots were cleaned. After being washed with de-ionized water and dried with absorbent paper, the plants were weighted with the electronic balance to get their fresh weight. Afterwards, plant organs were separated and put into envelopes to get dried at 70 °C. Dry weight of the various plant organs was then measured. Total Nitrogen (TN) and total Phosphorous (TP) contents of the dried plant organs were measured with the  $\text{H}_2\text{SO}_4\text{-H}_2\text{O}_2\text{-Colorimetric}$  method (Shi, 1992).

#### 2.3.3. Analysis of substrate samples

Substrates close to the plant roots were taken out to measure their TN and TP contents at both the beginning and finish of the experiments. Substrates were first dried at the room temperature and passed through the 100 mesh screen. 5 g of substrates were then added into the 150 mL conical flask. After adding 100 mL of de-ionized water, the flask was placed over the shaker at a rotational speed of 160 rpm for 48 h. The mixture in the flask was then filtered to measure TN and TP concentrations of the filtrate. The substrates in the control group without plants went through the same procedures to get their TN and TP contents. In addition, the activity of urease and phosphatase in the substrates was measured in the same manner as Kong et al. (2009).

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